

### **IN THE CLAIMS:**

1. (Currently amended) A method of detecting a difference of one or more nucleotides between a nucleic acid molecule to be tested and a reference nucleic acid molecule, said method comprising subjecting the test nucleic acid molecule to ~~base specific~~ single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments based on mass by MALDI-TOF MS ~~and/or~~ or other equivalent procedure to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure wherein the presence of an altered peak is indicative of a difference of one or more nucleotides in said tested nucleic acid molecule.
2. (Original) A method according to claim 1 wherein the nucleic acid molecule to be tested is amplified by a polymerase chain reaction (PCR) prior to base specific cleavage.
3. (Previously Amended) A method according to claim 1 wherein the base specific cleavage results in oligonucleotide fragments of from about 2 bases to about 1000 bases.
4. (Original) A method according to claim 3 wherein the base specific cleavage results in oligonucleotide fragments of from about 3 bases to about 500 bases.
5. (Original) A method according to claim 4 wherein the base specific cleavage results in oligonucleotide fragments of from about 4 bases to about 100 bases.
6. (Previously Amended) A method according to claim 1 wherein the base specific cleavage is uracil specific cleavage.
7. (Original) A method according to claim 6 wherein the uracil specific cleavage is mediated by uracil-N-glycosylase.

8. (Previously Amended) A method according to claim 1 further comprising subjecting fragmentation products to further separation (PSD) to generate a spectrum from decay dependent on the nucleotide sequence of the oligonucleotide.
9. (Original) A method according to claim 8 wherein the further separation of fragmentation products is by post source decay (PSD).
10. (Currently Amended) A computer program which controls a method of detecting a difference of one or more nucleotides between a nucleic acid molecule to be tested and a reference nucleic acid molecule, said method comprising subjecting the test nucleic acid molecule to ~~base specific~~ single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments based on mass by MALDI-TOF MS or other equivalent procedure to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure, wherein the presence of an altered peak is indicative of a difference of one or more nucleotides in said tested nucleic acid molecule.
11. (Original) A method according to claim 9 wherein the nucleic acid to be tested is amplified by PCR prior to base specific cleavage.
12. (Previously Amended) A method according to claim 9 wherein the base specific cleavage results in oligonucleotide fragments of from about 2 bases to about 1000 bases.
13. (Original) A method according to claim 9 wherein the base specific cleavage results in oligonucleotide fragments of from about 3 bases to about 500 bases.
14. (Original) A method according to claim 10 wherein the base specific cleavage results in oligonucleotide fragments of from about 4 bases to about 100.

15. (Previously Amended) A method according to claim 9 wherein the base specific cleavage is uracil specific cleavage.

16. (Original) A method according to claim 14 wherein the uracil specific cleavage is mediated by uracil-N-glycosylase.

17. (Previously Amended) A method according to claim 10 further comprising the further separation of fragmentation products to generate a spectrum form decay dependent on the nucleotide sequence of the oligonucleotide.

18. (Original) A method according to claim 17 wherein the further separation of fragmentation products is by post source decay (PSD).

19-23. (Previously cancelled)

24. (Currently Amended) A method for identifying or locating a mutation in one or more bases in a target nucleic acid molecule, comprising subjecting the target nucleic acid molecule to ~~base specific~~ single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments based on mass by MALDI-TOF MS or other equivalent procedure to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment, and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure, wherein the presence of an altered peak is indicative of a mutation in one or more bases in said target nucleic acid molecule.

25. (Original) A method according to claim 24 wherein the nucleic acid molecule to be tested is amplified by a polymerase chain reaction (PCR) prior to base specific cleavage.

26. (Previously Amended) A method according to claim 24 wherein the base specific cleavage results in oligonucleotide fragments of from about 2 bases to about 1000 bases.

27. (Original) A method according to claim 26 wherein the base specific cleavage results in oligonucleotide fragments of from about 3 bases to about 500 bases.
28. (Original) A method according to claim 27 wherein the base specific cleavage results in oligonucleotide fragments from about 4 bases to about 100 bases.
29. (Previously Amended) A method according to claim 24 wherein the base specific cleavage is uracil specific cleavage.
30. (Original) A method according to claim 29 wherein the uracil specific cleavage is mediated by uracil-N-glycosylase.
31. (Previously Amended) A method according to claim 24 further comprising subjecting fragmentation products to further separation (PSD) to generate a spectrum from decay dependent on the nucleotide sequence of the oligonucleotide.
32. (Original) A method according to claim 31 wherein the further separation of fragmentation products is by post source decay (PSD).
33. (New) A method of detecting a difference of one or more nucleotides between a nucleic acid molecule to be tested and a reference nucleic acid molecule, said method comprising subjecting the test molecule to single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments based on mass by MALDI-TOF MS or other equivalent procedure to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment, and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure, wherein the presence of an altered peak is indicative of said difference of one or more nucleotides, and wherein the difference does not result in a change of a cleavage site.

34. (New) A computer program which controls a method of detecting a difference of one or more nucleotides between a nucleic acid molecule to be tested and a reference nucleic acid molecule, said method comprising subjecting the test nucleic acid molecule to single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure, wherein the presence of an altered peak is indicative of a difference of one or more nucleotides in said tested nucleic acid molecule, and wherein the difference does not result in a change of a cleavage site.

35. (New) A method for identifying or locating a mutation in one or more bases in a target nucleic acid molecule wherein the mutation does not result in a change of a cleavage site by a restriction enzyme, comprising subjecting the target nucleic acid molecule to single-base-specific cleavage to generate oligonucleotide fragments, separating the resulting oligonucleotide fragments based on mass by MALDI-TOF MS or other equivalent procedure to produce a fingerprint of the oligonucleotide fragments comprising one or more peaks wherein a peak represents the mass of each fragment, and identifying an altered peak relative to a reference nucleic acid molecule subjected to the same procedure, wherein the presence of an altered peak is indicative of a mutation in one or more bases in said target nucleic acid molecule, and wherein the difference does not result in a change of a cleavage site.

## **REMARKS**

In the Office Action dated March 28, 2003, claims 1-18 and 24-32 are pending and are under consideration. Claims 1-5 and 24-28 are rejected under 35 U.S.C. §102(e) as allegedly anticipated by Kamb (U.S. Patent 5,869,242). Claims 6-7 and 29-30 are rejected under 35 U.S.C. §103(a) over Kamb in view of Sutherland et al. (U.S. Patent 5,985,619). Claims 10 and 14 are rejected under 35 U.S.C. §103(a) over Kamb in view of Koster (U.S. Patent 6,074,823). Claims 8-9, 11-13 and 31-32 are rejected under 35 U.S.C. §103(a) over Kamb in view of Caprioli (U.S. Patent 5,808,300). Claim 16 is rejected under 35 U.S.C. §103(a) over Kamb in view of Koster and further in the view of Sutherland et al. Claim 15 rejected under 35 U.S.C. §103(a) over Kamb in view of Caprioli and further in view of Sutherland et al. Claims 17-18 are rejected under 35 U.S.C. §103(a) over Kamb in view of Koster and further in view of Caprioli.

Applicants' representatives conducted a telephone interview with Examiner Chakrabarti (hereinafter "the Examiner") and Supervisory Examiner Benzion on July 9, 2003. Applicants thank the Examiners for the courtesy and assistance extended to Applicants during the telephone interview.

During the telephone interview, the Kamb reference (U.S. Patent 5,869,242) was discussed. The Examiner indicated that both the §102(e) and the §103(a) rejections would be withdrawn once the claims are properly amended to distinguish the method as disclosed by Kamb. More specifically, the Examiner suggested clarifying the term "base-specific cleavage", e.g., by reciting "single-base-specific cleavage", to distinguish cleavage by restriction endonuclease, which is allegedly disclosed by Kamb.

In an effort to expedite favorable prosecution and consistent with the Examiners recommendation, Applicants respectfully submit that independent claims 1, 10 and 24 have been

amended to substitute the term “base specific cleavage” with the term “single-base-specific cleavage”. Applicants respectfully submit that single-base-specific cleavage, i.e., cleavage at each occurrence of a certain base (e.g., at each A, each C, each G, each T or each U), is supported by the specification and is specifically exemplified by cleavage with uracil-N-glycosidase (which cleaves at each U) at pages 17-18 of the specification. No new matter is introduced by this amendment.

Applicants further respectfully submit that in contrast to the present invention, the Kamb reference merely teaches a detection method which employs restriction enzymes as cleavage agents. In the first instance, restriction enzymes act upon specific strings of sequences, i.e., cleavage by restriction enzymes is not specific to a single base. In addition, Kamb’s method employing restriction enzymes is limited to detecting mutations which occur at the cleavage site of a restriction enzyme.

Furthermore, Applicants respectfully submit that Kamb does not adequately teach a method of detecting mutations using a single-base-specific cleavage agent. It is recognized that Kamb mentions, in passing, the use of uracil-N-glycosidase at column 4, lines 39-41. There is no teaching in Kamb as to whether uracil-N-glycosidase alone can produce fragments which, upon separation based on mass, will produce a distinguishing fingerprint, as presently claimed, or whether uracil-N-glycosidase should be used in addition to a restriction enzyme. In addition, it is unclear based on Kamb as to whether a mutation, which does not affect the cleavage by uracil-N-glycosidase, can be detected. Therefore, Applicants respectfully submit that Kamb does not adequately teach a method of detecting a difference of one or more nucleotides between two nucleic acids by employing single-base-specific cleavage and separation of cleavage products based on mass, as presently claimed.

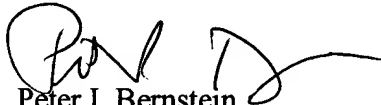
Accordingly, Applicants respectfully submit that the rejection under 35 U.S.C. §102(e) based on Kamb is overcome. Withdrawal of the rejection is respectfully requested.

Applicants further submit that none of the secondary references, which are relied upon in raising the §103 rejections, cure the deficiencies of the Kamb reference, as the Examiner apparently conceded during the telephone interview. Therefore, withdrawal of all rejections under 35 U.S.C. §103 is respectfully requested.

Applicants further submit that claims 33-35 are added to further delineate the claimed invention, i.e., that the difference of one or more nucleotides being detected does not result in a change of a cleavage site. Support for this amendment is found in original claims 1, 10 and 24, and in the specification, e.g., at page 19, lines 27-31 and Figure 1. No new matter is introduced.

In view of the foregoing amendments and remarks, it is firmly believed that the subject application is in condition for allowance, which action is earnestly solicited.

Respectfully submitted,

  
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